

Claims

1. (Withdrawn) An isolated protein, comprising an amino acid sequence having at least 80% sequence identity to SEQ ID NO: 2, wherein the protein has taxoid oxygenase activity.

2. (Canceled).

3. (Withdrawn) The isolated protein of claim 1, comprising an amino acid sequence as set forth in SEQ ID NO: 2.

4. (Canceled).

5. (Currently Amended) An isolated nucleic acid molecule encoding a protein comprising an amino acid sequence having at least 95% ~~80%~~ sequence identity to SEQ ID NO: 2, wherein the protein has taxoid oxygenase activity.

6. (Canceled).

7. (Currently Amended) An isolated nucleic acid molecule according to claim 5 comprising the sequence of as set forth in ~~SEQ ID NO: 1.~~

8. (Currently Amended) A recombinant nucleic acid molecule, comprising a promoter sequence operably linked to a the nucleic acid molecule according to claim 5.

9. (Currently Amended) A cell transformed with a the recombinant nucleic acid molecule according to claim 8.

10. (Original) The cell of claim 9, wherein the cell is a plant cell, an insect cell, a bacterium, or a yeast cell.

11-12. (Canceled).

13. (Withdrawn) A non-human transgenic organism, comprising a recombinant nucleic acid molecule according to claim 8.

14. (Withdrawn) The organism of claim 13, wherein the organism is a plant.

15-23. (Canceled).

24. (Withdrawn) A non-human transgenic organism, comprising a recombinant nucleic acid molecule according to claim 19.

25. (Withdrawn) The organism of claim 24, wherein the organism is a plant.

26. (Canceled).

27. (Currently Amended) An isolated nucleic acid molecule, comprising a sequence having at least ~~80%~~ 95% sequence identity with SEQ ID NO: 1, wherein the nucleic acid molecule encodes a protein having taxoid oxygenase activity.

28. (Canceled).

29. (Withdrawn) A taxoid oxygenase protein encoded by the nucleic acid molecule of claim 27.

30. (Original) A recombinant nucleic acid molecule, comprising a promoter sequence operably linked to the nucleic acid molecule of claim 27.

31. (Currently Amended) A cell transformed with ~~a~~ the recombinant nucleic acid molecule according to claim 30.

32. (Original) The cell of claim 31, wherein the cell is a plant cell, an insect cell, a bacterium, or a yeast cell.

33-34. (Canceled).

35. (Withdrawn) A non-human transgenic organism, comprising a recombinant nucleic acid molecule according to claim 30.

36. (Withdrawn) The organism of claim 35, wherein the organism is a plant.

37. (Canceled).

38. (Withdrawn) A method of identifying a nucleic acid sequence that encodes a taxoid oxygenase, comprising:

(a) hybridizing a probe to a first nucleic acid sequence under high stringency conditions, wherein the probe comprises at least 600 contiguous nucleotides of the nucleic acid molecule of claim 7; and

(b) determining that a protein encoded by the first nucleic acid sequence is capable of oxidizing a taxoid substrate, thereby identifying the first nucleic acid sequence as a nucleic acid sequence that encodes a taxoid oxygenase.

39-40. (Canceled).

41. (Withdrawn and Currently Amended) A method of hydroxylating a substrate, comprising:

~~contacting a cell substrate with at least one oxygenase comprising the isolated nucleic acid molecule according to claim 5 with a substrate encoding a protein comprising an amino acid sequence having at least 95% 80% sequence identity to SEQ ID NO: 2; and~~

allowing an oxygenase encoded by the isolated nucleic acid molecule to hydroxylate the substrate.

42. (Canceled).

43. (Withdrawn and Original) The method of claim 41, wherein the substrate is a taxoid.

44-46. (Canceled).

47. (Withdrawn and Original) The method of claim 43, wherein the oxygenase hydroxylates position C5 of the taxoid.

48. (Canceled).

49. (Withdrawn and Currently Amended) The method of claim 41, wherein the oxygenase is expressed in an isolated cell is selected from or in a transgenic plant cell, bacterium cell, insect cell, fungus cell or yeast cell, and the hydroxylation of the substrate occurs *in vivo*.

50. (Withdrawn and Currently Amended) The method of claim 41, wherein the substrate is an exogenous substrate, ~~which is fed to the isolated cell, transgenic plant, transgenic bacterium, transgenic insect, transgenic fungus or transgenic yeast.~~

51. (Withdrawn and Currently Amended) A method for increasing paclitaxel yield in a cell, comprising introducing the nucleic acid molecule according to claim 5 ~~at least one nucleic acid molecule of claim 27~~ into a paclitaxel-producing cell, wherein an amount of paclitaxel produced by the cell increases following introduction of the ~~at least one~~ nucleic acid molecule into the cell.

52. (Withdrawn and Currently Amended) The method of claim 51, wherein introducing the ~~at least one~~ nucleic acid molecule into the cell comprises transfecting the cell with the ~~at least one~~ nucleic acid molecule.

53. (Withdrawn and Currently Amended) The method of claim 51, wherein the ~~at least one~~ nucleic acid molecule comprises a ~~sequence as set forth in the~~ protein-coding region of SEQ ID NO: 1.

54. (Canceled).

55. (Withdrawn and Original) The method of claim 51, wherein the cell is a *Taxus* cell.

56. (Withdrawn and Original) The method of claim 51, further comprising introducing into the cell an additional nucleic acid molecule selected from the group consisting of:

(a) a first nucleic acid sequence encoding a protein having taxadiene synthase activity;

(b) a second nucleic acid sequence encoding a protein having taxadien-5-ol transacylase activity;

(c) a third nucleic acid molecule encoding a protein having taxadien-2-ol transacylase activity;

(d) one or more fourth nucleic acid molecules encoding one or more proteins having taxoid oxygenase activity;

(e) a fifth nucleic acid molecule encoding a protein having 10-deacetylbaecatin III-10-O-acetyl transferase activity;

(f) a sixth nucleic acid molecule encoding a protein having taxoid 13-phenylpropanoyltransferase activity;

(g) a seventh nucleic acid molecule encoding a protein having 3'-N-debenzoyltaxol N-benzoyltransferase activity; and

(h) combinations of (a), (b), (c), (d), (e), (f), or (g).

57. (Withdrawn and Original) The method of claim 56, wherein:

(a) the first nucleic acid molecule comprises a nucleic acid sequence having at least 90% sequence identity to the protein-coding region of SEQ ID NO: 19 and encodes a protein having taxadiene synthase activity;

(b) the second nucleic acid molecule comprises a nucleic acid sequence having at least 90% sequence identity to the protein-coding region of SEQ ID NO: 21 and encodes a protein having taxadien-5-ol transacylase activity;

(c) the third nucleic acid molecule comprises a nucleic acid sequence having 90% sequence identity to the protein-coding region of SEQ ID NO: 23 and encodes a protein having taxadien-2-ol transacylase activity;

(d) the one or more fourth nucleic acid molecules comprise a nucleic acid sequence having 90% sequence identity to any one of the sequences set forth in SEQ ID NOs: 3, 5, 7, 9, 11, 13, 15, 17, and 40 and encode a protein having taxoid oxygenase activity;

(e) the fifth nucleic acid molecule comprises a nucleic acid sequence having 90% sequence identity to the protein-coding region of SEQ ID NO: 34 and encodes a protein having 10-deacetylbaccatin III-10-O-acetyl transferase activity;

(f) the sixth nucleic acid molecule comprises a nucleic acid sequence having 90% sequence identity to the protein-coding region of SEQ ID NO: 36 and encodes a protein having taxoid 13-phenylpropanoyltransferase activity; and

(g) the seventh nucleic acid molecule comprises a nucleic acid sequence having 90% sequence identity to the protein-coding region of SEQ ID NO: 38 and encodes a protein having 3'-N-debenzoyltaxol N-benzoyltransferase activity.

58. (Withdrawn and Original) The method of claim 56, wherein the one or more fourth nucleic acid molecules encode one or more proteins having taxoid 7 β -hydroxylase activity, taxoid 14 β -hydroxylase activity, taxoid 10 β -hydroxylase activity or taxoid 13 α -hydroxylase activity.

59. (Canceled).

60. (Withdrawn and Original) The method of claim 56, wherein the additional nucleic acid molecule is selected from the group consisting of:

(a) a first nucleic acid molecule comprising a nucleic acid sequence having at least 90% sequence identity to the protein-coding region of SEQ ID NO: 19, wherein the first nucleic acid sequence encodes a protein having taxadiene synthase activity;

(b) a second nucleic acid molecule comprising a nucleic acid sequence having at least 90% sequence identity to the protein-coding region of SEQ ID NO: 21, wherein the second nucleic acid sequence encodes a protein having taxadien-5-ol transacylase activity;

(c) a third nucleic acid molecule comprising a nucleic acid sequence having 90% sequence identity to the protein-coding region of SEQ ID NO: 23, wherein the third nucleic acid sequence encodes a protein having taxadien-2-ol transacylase activity; and

(d) combinations of (a), (b), or (c).

61-62. (Canceled).

63. (Withdrawn) An antibody or antibody fragment that binds the protein of claim 1.

64-65. (Canceled).